

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

LISTING OF CLAIMS:

1. (currently amended): A method for assaying concentrations of Coenzyme A molecules in a biological samples, ~~the said method being characterized by~~ comprising a step of extraction from ~~a said~~ biological sample using a strongly acidic solution, a step of solid phase extraction, a step of adding an internal standard substance, and a step of detection by LC-MS, wherein said internal standard substance is not present in said biological sample prior to said extraction using a strongly acidic solution.
2. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 1, ~~characterized in that~~ wherein the step of extracting the Coenzyme A molecules from the biological sample using a strongly acidic solution comprises freezing and pulverizing said ~~is a step wherein a freeze-shattered biological sample, mixing said frozen and pulverized biological sample with~~ is agitated in a perchloric acid solution to form a suspension, and subjecting said suspension ~~the supernatant is subjected to~~ centrifugal separation.
3. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 1 or 2, ~~characterized in that~~ wherein ~~the said~~ solid phase extraction step is a step wherein the supernatant obtained by extraction of the Coenzyme A molecules with a strongly acidic solution is neutralized, and then applied to a reverse phase cartridge packed with silica gel containing an octadecylsilyl group or octylsilyl group, washed with an aqueous solvent, and eluted with an organic solvent.

4. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 3, ~~characterized in that~~wherein the supernatant is applied to said reverse phase cartridge after conditioning ~~the of said~~ reverse phase cartridge with acetonitrile and 1 M ammonium acetate solution.

5. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 3, ~~characterized in that~~wherein ~~the said~~ organic solvent is a mixture of acetonitrile and ammonium acetate.

6. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 1, ~~characterized in that~~wherein the Coenzyme A molecules ~~is~~are fatty acid Coenzyme A esters, and ~~the said~~ internal standard substance is a structural analog of the Coenzyme A molecules.

7. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 6, ~~characterized in that~~wherein the fatty acid Coenzyme A esters ~~is~~are a Coenzyme A esters of a short chain fatty acid with 2-8 carbons in the main carbon chain, and the structural analog has a difference of no more than 3 carbons with the Coenzyme A molecule and has at least 3 of the hydrogens of the main chain substituted with deuterium, or has at least 3 of the carbons of the main carbon chain substituted with ^{13}C .

8. (currently amended): The method for assaying concentrations of Coenzyme A molecules according to claim 7, ~~characterized in that~~wherein the Coenzyme A molecules ~~is~~are malonyl CoA and the structural analog is acetyl CoA-d₃, methylmalonyl CoA-d₃, methylmalonyl CoA-d₄, propionyl CoA-d₃, propionyl CoA-d₅ or malonyl CoA- $^{13}\text{C}_3$.